

**HEK 293**

# Optimum Growth™ 5L Flasks:

## High and Low Expressing Proteins in HEK 293 Cells

Data provided by:

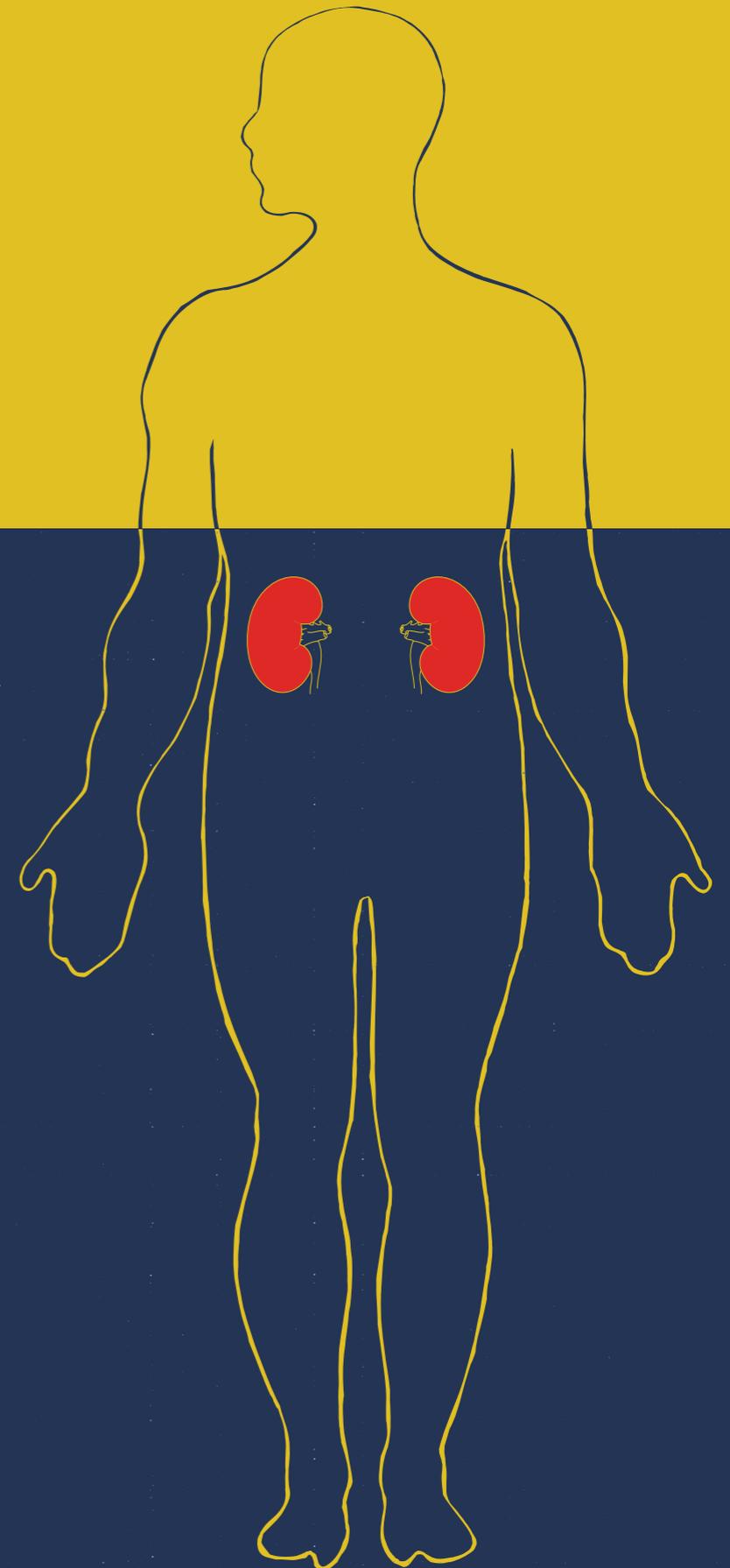


## Consistent and Improved Expression

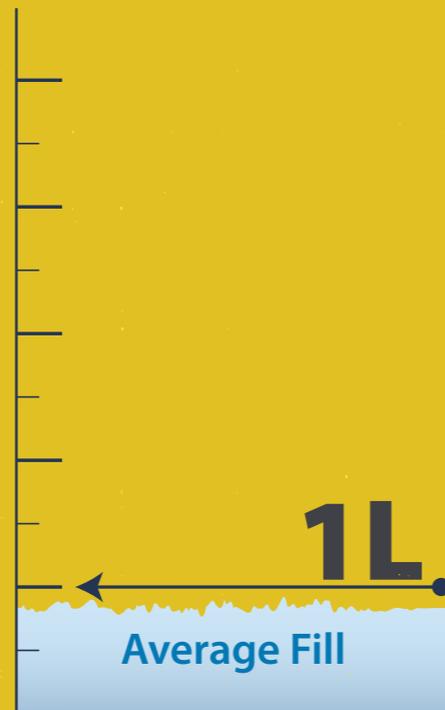
Thomson Optimum Growth 5L Flasks (patented) produce consistent expression across replicates and production runs for both high and low expressing proteins in HEK 293 cells. This is particularly important when working with constructs on the low end, when fluctuations can lead to unacceptable expression levels. Optimum Growth 5L flasks are able to maintain constant expression often elevated levels compared to other culture flasks. NGM Biopharmaceuticals uses the Optimum Growth™ 5L flasks to get greater volume and yield from the same footprint as Corning® 3l flasks.

# The Following Data Represents How Thomson 5l Flasks Can Aid Your Production on Both End of the Expression Spectrum.

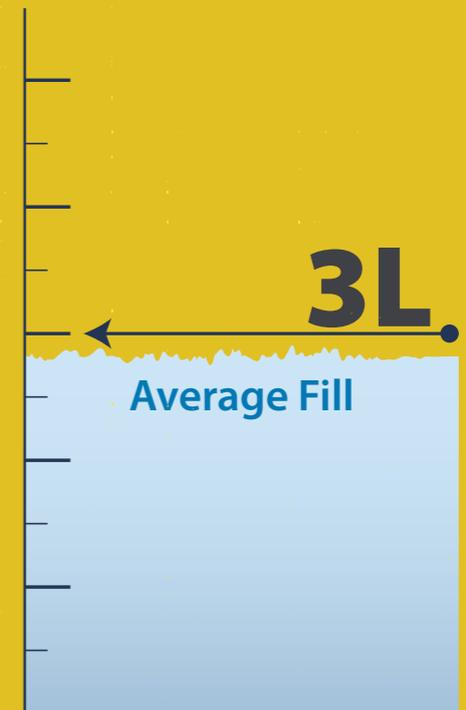
- All data provided represent transient expression data using HEK293 cells using a harvest schedule of 6-7 days post-transfection
- Transfections were performed at the 2L scale with replicates to check for reproducibility and consistency.
- The 5L flasks were maintained in a reach in CO2 incubator maintained at 37C and 5% CO2 level.
- The flasks were maintained on an Innova 2300 platform shaker set at 80 RPM (for a 2 inch radius).



# Fill Volumes



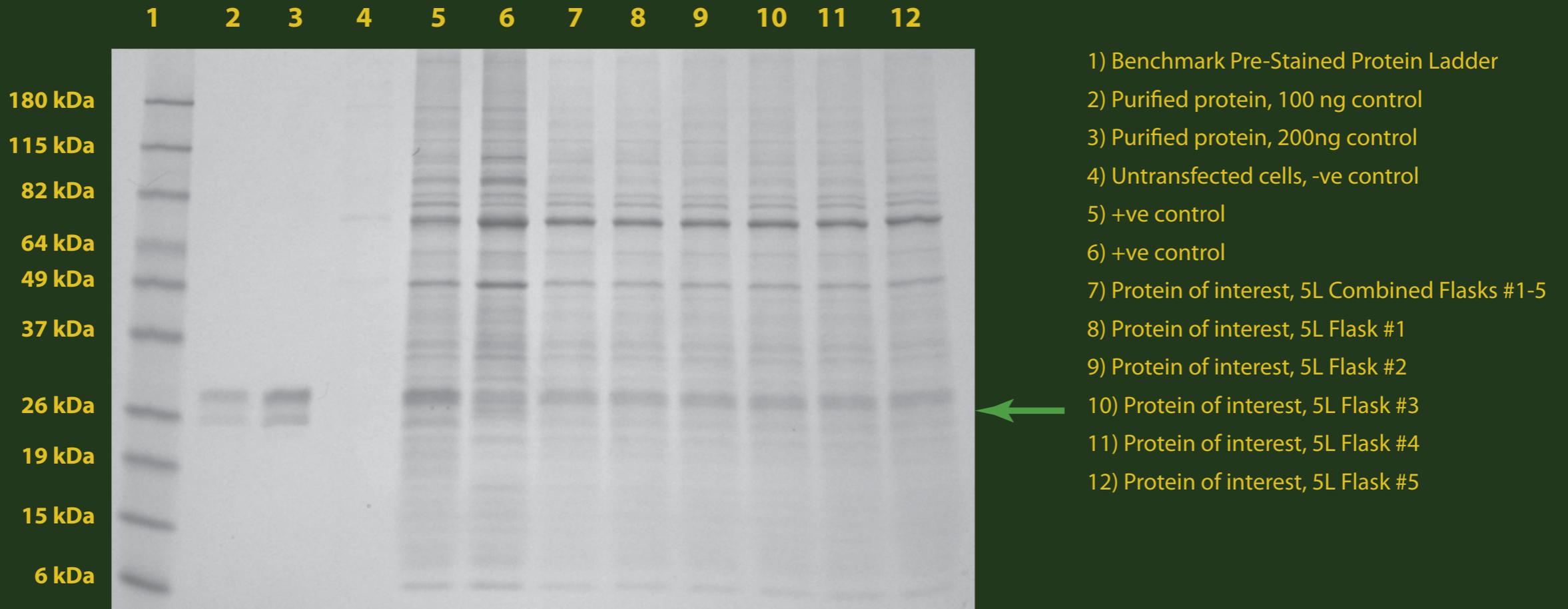
**Corning® 3L Flask**  
*(1L Working Volume)*



**Optimum Growth™ 5L Flask**  
*(3L Working Volume)*

# LOW EXPRESSING GEL

This gel shows equal bands from 5 replicates of a low expressing protein, producing roughly 10 to 20 mg/L.



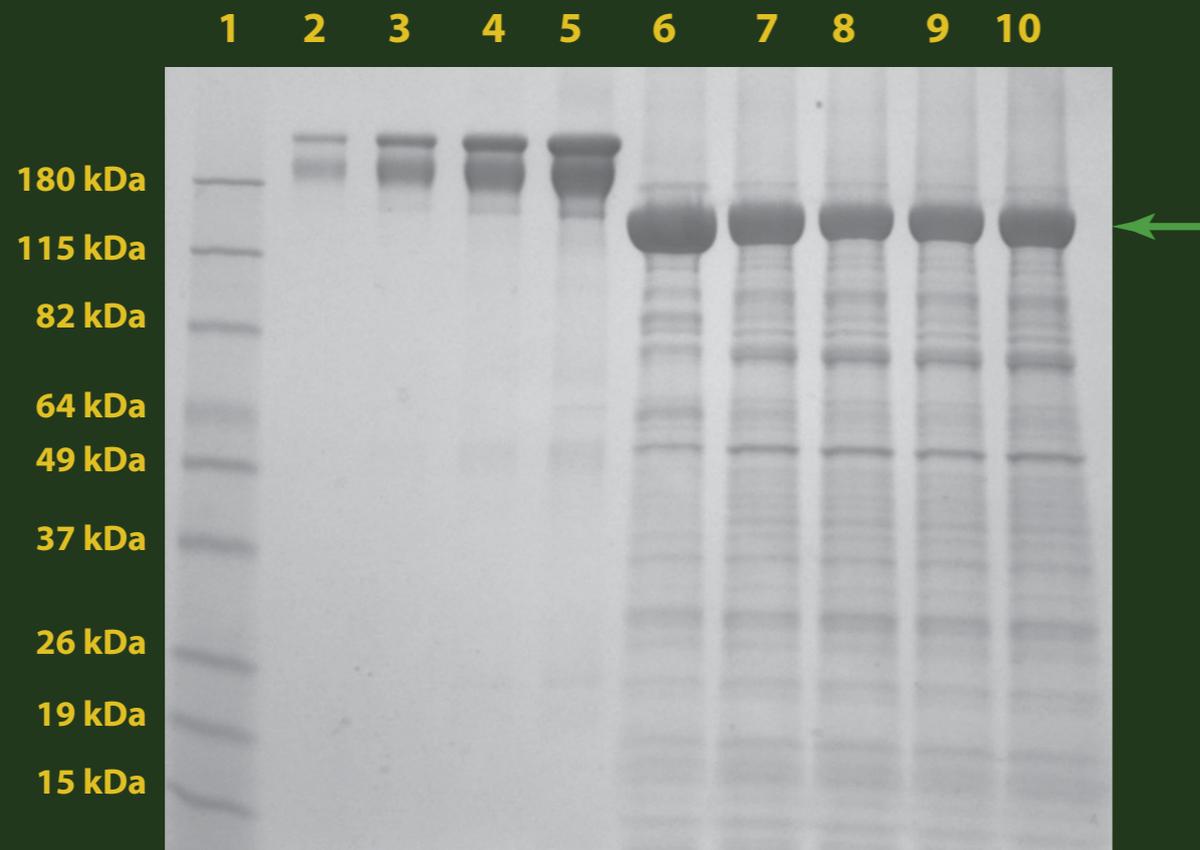
4-20% SDS-PAGE Quick Blue Stain Commassie Gel

Expected MW of dimer 24.5 kDa

Estimated expression level ~10-20 mg/L

# HIGH EXPRESSING GEL

Thomson 5L flasks are also able to consistently maximize production of your best expressers. This gel shows equal bands from 3 replicates of a high expressing protein, producing roughly 300 mg/L.



4-20% SDS-PAGE Quick Blue Stain Commassie Gel

- 1) Benchmark Pre-Stained Protein Ladder
- 2) Purified mAb 100 ng control
- 3) Purified mAb 250 ng control
- 4) Purified mAb 500 ng control
- 5) Purified mAb 1000 ng control
- 6) +ve control
- 7) Protein of interest, 5L Flask #1
- 8) Protein of interest, 5L Flask #2
- 9) Protein of interest, 5L Flask #3
- 10) Protein of interest, 5L Combined Flasks #1-3

Expected MW of dimer 159.4 kDa    Estimated expression level ~300 mg/L

# Conclusion

Thomson Optimum Growth flasks not only ensure consistent expression from your Hek293 strains, they can also increase shaker capacity. With the same footprint as a typical Corning® 3L flask and a culture volume of up to 3L, the 5L flask may increase production 200%, if not more, in the same space (this is construct dependent).

Most constructs express at higher levels in the Optimum Growth 5L flasks. This makes one Optimum Growth 5L equivalent to, if not greater than, two 3L flasks.



**Corning® 3L Flask**  
*(1L Working Volume)*

**Corning® 3L Flask**  
*(1L Working Volume)*

**Optimum Growth™ 5L Flask**  
*(3L Working Volume)*